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## Insulin uptake and effect on glucose utilization by ovine and bovine adipose tissue cultured over 7 days

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maximal steatosis as early as week 2 (168 and 292 mg/10<sup>9</sup> cells, respectively). Our results showed that NEFA is the best plasma predictor for TG liver infiltration in weeks 1 ( $r = 0.68$ ), 2 ( $r = 0.42$ ) and 4 ( $r = 0.57$ ). Furthermore, plasma LDL, apo B and A-I are other potential predictors with the accuracy of prediction dependent on the time post-partum.

**Insulin uptake and effect on glucose utilization by ovine and bovine adipose tissue cultured over 7 days.** Y Faulconnier, L Guillon, R Lefavre, M Tourret, Y Chilliard (*INRA, Laboratoire Sous-Nutrition des Ruminants, 63122 Saint-Genès-Champanelle, France*)

The effect of insulin (2 mU/ml) on glucose utilization was studied on adipose tissue (AT) explants from non-lactating non-pregnant cows ( $n = 5$ ) and ewes ( $n = 5$ ) fed a restricted diet (20–22% of energy maintenance requirement, EMR) for 8–10 d and then overfed (188 or 228% of EMR, for cows or ewes) during 21 (cows) or 10 (ewes) d until slaughter, to induce a rebound in lipogenic activities. The body condition (scale 0–5) of the cows and ewes averaged  $2.5 \pm 0.6$  and  $3.0 \pm 0.3$ , and the mean adipocyte diameter  $122 \pm 3$  and  $111 \pm 9 \mu\text{m}$ , respectively.

Samples of perirenal AT were cut into 10–15 mg pieces, and cultured over 7 d in sterile conditions in medium 199 supplemented with acetate (7.0 mM). The culture medium was changed daily. Glucose concentration was measured using the glucose dehydrogenase method. The loss of insulin from the cultured medium in the 2 species ranged from 60 (1st d of culture) to approximately 35% (d 2 to d 7) of the initial amount.

In basal conditions, the glucose utilization was similar (33–24 and 16  $\mu\text{mol}/24 \text{ h}/10^6$  adipocytes on d 1–2, 3–4 and 5–6–7, respectively) in the 2 species. However, the glucose utilization by bovine AT was higher (+18%) during the 2nd than during the 1st d, in contrast to ovine AT where this utilization decreased (–13%) during this period. In the 2 species, the glucose utilization then progressively decreased until d 7.

The addition of insulin increased ( $P < 0.001$ ) glucose utilization in the 2 species. However, the effect of insulin was different according to the species and the day of culture (interaction species x insulin x day,  $P < 0.03$ ). The effect of insulin on

d 1 was greater in bovine (+67%) than in ovine (+20%) AT, whereas on d 3–4 and d 5–6–7 this effect was greater in ovine (+92 and 132%) than in bovine (+64 and 50%) AT.

This study indicates that ruminant AT explants remain metabolically active for at least 7 d when maintained in a suitable medium, with interactions between insulin and animal species that affect glucose utilization.

**Hepatic apo B and mRNA apo B levels in the underfed high-producing dairy cow during early lactation.** D Gruffat<sup>1</sup>, F Duboisset<sup>1</sup>, D Durand<sup>1</sup>, J Lefavre<sup>1</sup>, A Ollier<sup>2</sup>, G d'Onofrio<sup>3</sup>, P Williams<sup>4</sup>, Y Chilliard<sup>2</sup>, D Bauchart<sup>1</sup> (<sup>1</sup> *INRA, Laboratoire Croissance et Métabolismes des Herbivores*; <sup>2</sup> *INRA, Laboratoire Sous-Nutrition des Ruminants, 63122 Saint-Genès-Champanelle*; <sup>3</sup> *INSERM U 321, Hôpital de la Pitié, 75013 Paris*; <sup>4</sup> *Rhône Poulenc Nutrition Animale, 03600 Commentry, France*)

In high-producing dairy cows in early lactation, intense mobilization of lipids is often associated with fatty liver syndrome. The limited capacity of the liver to export triglyceride-rich lipoproteins in the form of very low density lipoproteins (VLDL) increases the risk of development of fatty liver. In cows in early lactation, synthesis of apolipoprotein B (apo B), the major apolipoprotein in VLDL, may be a rate-limiting step for hepatic VLDL production and secretion.

Eight multiparous H x F fat cows after calving (body condition score: 3.9/5) were offered a low concentrate (< 25% DM diet) – corn silage complete diet to increase fat mobilization and to induce hepatic steatosis. All cows were liver biopsied (400 mg/cow; samples frozen in liquid nitrogen) at 1, 2, 4, and 12 weeks after calving. Total hepatic RNA was extracted from each biopsy sample using guanidium thiocyanate/phenol/chloroform. Levels of mRNA for apo B were determined by the Dot-blot method using a cDNA human probe and hepatic apo B levels were measured by Western-blot using rabbit specific antibody against bovine apo B. Hepatic DNA was determined by fluorimetric method and was used to express the apo B levels per 10<sup>9</sup> hepatic cells.

Levels of mRNA of apo B remained stable after calving ( $8.4 \pm 1.0$ ,  $8.3 \pm 1.8$ ,  $10.6 \pm 1.3$ , and